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Effect of a multistage ultraendurance triathlon on aldosterone, vasopressin, extracellular water and urine electrolytes

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Abstract: Prolonged endurance exercise over several days induces increase in extracellular water (ECW). We aimed to investigate an association between the increase in ECW and the change in aldosterone and vasopressin in a multistage ultraendurance triathlon, the 'World Challenge Deca Iron Triathlon' with 10 Ironman triathlons within 10 days. Before and after each Ironman, body mass, ECW, urinary [Na(+)], urinary [K(+)], urinary specific gravity, urinary osmolality and aldosterone and vasopressin in plasma were measured. The 11 finishers completed the total distance of 38 km swimming, 1800 km cycling and 422 km running within 145.5 (18.8) hours and 25 (22) minutes. ECW increased by 0.9 (1.1) L from 14.6 (1.5) L prerace to 15.5 (1.9) L postrace ($P < 0.0001$). Aldosterone increased from 70.8 (104.5) pg/mL to 102.6 (104.6) pg/mL ($P = 0.033$); vasopressin remained unchanged. The increase in ECW was related neither to postrace aldosterone nor to postrace vasopressin. In conclusion, ECW and aldosterone increased after this multistage ultraendurance triathlon, but vasopressin did not. The increase in ECW and the increase in aldosterone were not associated.

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The effect of a multi-stage ultra-endurance triathlon on aldosterone, vasopressin, extracellular water and urine electrolytes – The World Challenge Deca Iron Triathlon 2009

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Abstract

Background and Aims: Prolonged endurance exercise over several days induces increases of extracellular water (ECW). We aimed to investigate an association between the increase in ECW and the change in aldosterone and vasopressin in a multi-stage ultra-endurance triathlon, the 'World Challenge Deca Iron Triathlon' with 10 Ironman triathlons within 10 days. **Methods:** Before and after each Ironman, body mass, ECW, urinary $[Na^+]$, urinary $[K^+]$, urinary specific gravity urinary osmolality and aldosterone and vasopressin in plasma were measured. **Results:** The 11 finishers completed the total distance of 38 km swimming, 1,800 km cycling and 422 km running within 145.5 (18.8) h and 25 (22) min. ECW increased by 0.9 (1.1) L from 14.6 (1.5) L pre race to 15.5 (1.9) L post race ($p < 0.0001$). Aldosterone increased from 70.8 (104.5) pg/mL to 102.6 (104.6) pg/mL ($p = 0.033$), vasopressin remained unchanged. The increase in ECW was neither related to post race aldosterone nor to post race vasopressin. The changes in aldosterone and vasopressin were not related to the increase in ECW. **Conclusions:** ECW and aldosterone increased after this multi-stage ultra-endurance triathlon, but not vasopressin. The increase in ECW and the increase in aldosterone were not associated.

Key words: body mass – dehydration – fluid – sodium – potassium

Introduction

Long-distance triathlons such as the Ironman triathlon are highly popular and are held since the first event in 1978 in Hawaii.¹ In 2006, for the first time, a multi-stage long-distance triathlon was held in Mexico, where athletes had to complete one Ironman distance per day for 10 days.²

In single Ironman triathlons, exercise-associated hyponatremia (EAH) is considered a frequent and serious problem.^{3,4} EAH is defined as a decrease in plasma $[\text{Na}^+]$ concentration $< 135 \text{ mmol/l}$ ⁵ and fluid overload is the likely aetiology.^{4,6} However, regarding recent findings, an increased activity of vasopressin may contribute to the pathogenesis of EAH in runners.^{7,8}

An interesting finding in multi-stage ultra-endurance performances is the increase in total body water.^{9,10} It has been well documented that prolonged strenuous exercise over several consecutive days induced a progressive increase in extracellular water, plasma volume and total body water.¹¹⁻¹⁵ Sodium retention seemed to be the major factor in the increase in plasma volume.⁹ In multi-stage ultra-endurance runs, plasma volume increased,¹³⁻¹⁵ plasma $[\text{Na}^+]$ concentration was maintained^{15,16} and aldosterone increased.^{13,14} Presumably fluid homeostasis during an ultra-endurance performance is regulated by both aldosterone and vasopressin.

Also for Ironman triathletes, an increase in plasma volume has been described.^{4,17} Ironman triathletes with symptomatic EAH showed abnormal fluid retention with an increased extracellular volume.⁴ Fluid retention, however, was not associated with an increased activity of vasopressin in Ironman triathletes.⁴

The association of vasopressin with effect on renal function and association with EAH has been investigated in marathon runners.¹⁸ Marathon runners developing EAH showed an increase in body mass^{19,20} and it was assumed that inappropriate anti-diuretic hormone release during exercise altered renal function²⁰ which may lead to water retention and EAH²¹.

The aim of this study was to investigate the association between the increase in both total body water and extracellular water with changes in both aldosterone and vasopressin during a multi-stage ultra-endurance triathlon, in order to determine whether the increase in total body water and extracellular water would be related to an increased activity in aldosterone or in vasopressin. We hypothesised that both vasopressin and aldosterone would be increased after a multi-stage ultra-endurance race, and that rather the increase in aldosterone would be related to the increase in both total body water and extracellular water than the increase in vasopressin.

Materials and Methods

Subjects

The organiser of the 'World Challenge Deca Iron Triathlon' in the 2009 edition contacted all participants via a asking them to participate in the study. The study was approved by the Ethical Committee of St. Gallen, Switzerland. All participants were informed about the experimental procedures and gave their informed written consent. All 17 participating athletes were interested and took part in the investigation. Of the 17 starters, 11 athletes with (mean and SD) 44.0 (10.5) years, 76.0 (7.3) kg body mass, 1.73 (0.06) m body height and a body mass index of 25.4 (2.0) kg/m² finished the race and were included in the study.

The race

The 'World Challenge Deca Iron Triathlon' was held in 2009 for the third time. The competition took place in the City of Monterrey in the Province of Nuevo León in northern Mexico. The race started on 9 November 2009. Seventeen male, and one female, ultra-endurance triathletes entered the race. Every day they had to complete the distance of one Ironman triathlon of 3.8 km swimming, 180 km cycling and 42.195 km running with a time limit of 24 hours. Every morning at 09:00 the event started with the swim in the 25 m indoor pool at the 'Centro Acuatico Olimpico Universitario' of 'Universidad Autonoma de Nuevo León' in Monterrey. The pool was heated and held at a constant temperature of 28 °Celsius. Laps of 25 m were counted by personal lap counters. After completing the swim, the athletes changed in the transition area and cycled to the 'Parque Niños Héroes'. This park was closed to traffic, completely illuminated and had a cycling track that was 95% flat, but included an inclination of 5%. The cycling consisted of 94 laps of 1.915 km each. After changing for the running course, the athletes first had to run a short lap of 703 m and then 22 laps of 1.886 km. In total, the athletes had to climb an altitude of 1,650 m per Ironman triathlon. Drafting during

cycling was strictly prohibited. Laps in the cycling and running course were counted electronically with a chip system. Athletes could be helped by their own support crew for nutrition and changes of equipment and clothes. During the whole race, accommodation was offered in the 'Parque Niños Héroes'. The athletes and their support crew had a room with bed, toilet and shower. For nutrition, the organiser offered a variety of food in a restaurant that was open 24 h.

Analysis of body composition

Before and after each stage, body composition was measured using the InBody 3.0 body composition analyser with direct segmental multifrequency bioelectrical impedance method (InBody 3.0, Biospace, Seoul, Korea) following Bedogni *et al.*²² **This method is reliable for determination of body water under both laboratory situations²² and field conditions.⁹** **However, hydration status must be unchanged²³ and extracellular osmolality must be stable²⁴.** InBody 3.0 has a tetrapolar 8-point tactile electrode system and performs 20 impedance measurements at each session, using four different frequencies (5 kHz, 50 kHz, 250 kHz, and 500 kHz) at each of five segments (right arm, left arm, trunk, right leg, and left leg). The subjects stood barefoot in an upright position on foot electrodes, with the legs and thighs not touching and the arms not touching the torso. Four foot electrodes were used and the subjects were asked to grip the two palm-and-thumb electrodes. They did this without shoes or excess clothing. The skin and electrodes were cleaned and dried before testing. Body mass, lean body mass, total body water, intracellular water, extracellular water, fat mass, and protein mass were directly determined.

Laboratory analysis

Before the start of the race, aldosterone and vasopressin were determined at rest. After venipuncture of an antecubital vein, the blood was centrifuged at 3,000 g for 10 min at 4

°Celsius; the serum was then separated, stored on ice, and then processed to determine both aldosterone and vasopressin using a radioimmunoassay method. The radioimmunoassay method measured quantitatively the concentration of aldosterone in the blood within a reference range of 10.0 pg/mL to 300 pg/mL, the activity of vasopressin within a reference range of 1.0 μ U/mL to 2.0 μ U/mL. The same procedure was repeated immediately upon arrival at the finish line after the last Stage on Day 10. Immediately before and immediately after each stage, samples of urine were collected. In the urine samples, $[\text{Na}^+]$, $[\text{K}^+]$, specific gravity and osmolality were determined. $[\text{Na}^+]$ and $[\text{K}^+]$ were determined using an ion selective electrode method. Specific gravity was measured manually using a NE Uricon refractometer which has a scale from 1,000 g/mL to 1,050 g/mL. To determine osmolality, urea was measured in the urine samples using a HITACHI 911 automated equipment using an enzymatic method; the formula urinary osmolality = $(\text{Na} + \text{K} \times 2) + \text{urea}/5.6$ was used to calculate osmolality with a reference range of 300 mosm/kg to 1,000 mosm/kg.

Statistical analysis

For finishers and non-finishers, overall changes of the variables over time were tested by MANOVA-procedure. In case of a significant change during the race paired t-tests were applied to detect a significant change from one time period to the other. For non-finishers, paired-t-tests were applied to detect changes from one time period to the other as long as the athlete was in the race and had completed a full stage. For finishers, Pearson correlation analysis was used to check for associations between parameters with statistically significant changes. Statistical significance was accepted with $p < 0.05$.

Results

Performance

A total of 17 male competitors started on 9 November 2009; six athletes did not reach the finish line. The first non-finisher dropped out on day 1 due to an accident, the second non-finisher stopped after day 2 due to overuse injuries of the lower limbs. After day 3, the third non-finisher stopped due to loss of motivation and the fourth non-finisher withdrew due to fatigue and overuse injuries of the lower limbs. On day 4, the fifth non-finisher had to stop due to respiratory problems and on day 5, the sixth non-finisher resigned due to fatigue and overuse injuries of the lower limbs. The 11 finishers completed the total distance of 38 km swimming, 1,800 km cycling and 422 km running within 145.5 (18.8) h and 25 (22) min, equal to 8,757 (1,129) min. **Table 1** represents the daily total race times per Ironman distance and the daily highest temperature. The time per Ironman increased throughout the race ($p < 0.0001$) (see **Figure 1**); the daily highest temperature was not related to the daily race time.

Changes in body composition during the race

For finishers, body mass decreased by 1.7 (2.4) kg from 76.3 (7.7) kg pre race to 74.6 (7.1) kg post race ($p < 0.0001$) (see **Figure 2**). Fat mass decreased by 3.2 (1.6) kg from 13.1 (4.0) kg pre race to 10.1 (3.6) kg post race ($p < 0.0001$). The decrease in body mass was significantly and positively related to the decrease in fat mass ($r = 0.71$, $p = 0.022$). Race time was neither correlated to pre race body mass nor to pre race fat mass. Lean body mass increased by 1.1 (2.5) kg from 62.9 (4.6) kg pre race to 64.0 (6.5) kg post race ($p < 0.01$). Protein mass showed no changes. Total body water increased by 1.5 (1.6) L from 46.3 (3.5) L pre race to 47.8 (4.1) L post race ($p < 0.0001$) (see **Figure 3**). Extracellular water increased by 0.9 (1.1) L from 14.6 (1.5) L pre race to 15.5 (1.9) L post race ($p < 0.0001$). Intracellular water showed no changes. The increase in total body water was related to the increase in extracellular water (r

= 0.64, $p = 0.047$). The increase in lean body mass was neither correlated to the increase in total body water ($r = -0.36$, $p = 0.33$) nor to the increase in extracellular water ($r = -0.36$, $p = 0.33$). Post race lean body mass was highly significantly correlated to both post race total body water ($r = 0.94$, $p < 0.0001$) and to post race extracellular water ($r = 0.81$, $p = 0.0044$).

Changes in urinary parameters, aldosterone and vasopressin

Urinary $[Na^+]$ showed no changes (see **Figure 4**), urinary $[K^+]$ and the potassium-to-sodium ratio in urine increased ($p < 0.0001$) for finishers. Urinary specific gravity and urinary osmolality showed no changes. Aldosterone increased from 70.8 (104.5) pg/mL pre race to 102.6 (104.6) pg/mL post race ($p = 0.033$); vasopressin remained unchanged at 1.34 (0.46) μ U/mL pre race to 1.68 (0.73) μ U/mL post race ($p = 0.252$). The change in aldosterone and the change in and the potassium-to-sodium ratio in urine were not associated ($r = -0.40$, $p = 0.28$), also post race aldosterone and post race potassium-to-sodium ratio in urine showed no association ($r = 0.05$, $p = 0.89$). The change in vasopressin was not related to the change in urinary osmolality ($r = -0.22$, $p = 0.59$) and post race vasopressin was not associated to post race urinary osmolality ($r = 0.10$, $p = 0.80$). The increase in extracellular water was neither related to post race aldosterone ($r = -0.36$, $p = 0.33$) nor to post race vasopressin ($r = -0.33$, $p = 0.38$). The changes in aldosterone ($r = -0.32$, $p = 0.40$) and vasopressin ($r = -0.28$, $p = 0.47$) were not related to the increase in extracellular water. Post race extracellular water was neither related to post race aldosterone ($r = -0.40$, $p = 0.28$) nor to post race vasopressin ($r = -0.26$, $p = 0.49$).

Results of the non-finishers

Table 2 represents the results of the non-finishers. After three stages, four non-finishers were still in the race. Body mass and fat mass decreased significantly whereas total body water and extracellular water showed no changes. While urinary sodium and urinary potassium

remained stable, the potassium-to-sodium ratio increased. Urinary specific gravity and urinary osmolality showed no changes.

Discussion

The aim of this study was to investigate the association between the increase in both total body water and extracellular water with changes in both aldosterone and vasopressin in a multi-stage ultra-endurance triathlon, in order to determine whether the increase in total body water and extracellular water would be related to an increased activity in aldosterone or in vasopressin.

A first hypothesis was that aldosterone would be increased after this multi-stage ultra-endurance triathlon. In line with our hypothesis, serum aldosterone increased significantly from 70.8 (104.5) ng/dL pre race to 102.6 (104.6) ng/dL post race. We see the increase in the potassium-to-sodium ratio as being due to an increased activity of aldosterone. The potassium-to-sodium ratio in urine was < 1.0 before the start of the race and increased to > 1.0 after Day 1 and before and after Day 2. This suggests that during this time more potassium than sodium was excreted through the kidney, and a positive ratio for potassium-to-sodium in urine suggests an increased activity of aldosterone. We interpret the increase in urinary potassium as a reaction to a stimulation of the renin-angiotensin-aldosterone-system (RAAS). This suggests that during these days more potassium than sodium was excreted through the kidneys and a positive ratio for potassium to sodium in urine suggests an increased activity of aldosterone. A potassium-to-sodium ratio in urine > 1.0 reflects a contraction of the effective extra-cellular volume leading to a hyperreninemic hyperaldosteronemia. The potassium-to-sodium ratio in urine is a physiological reflection of the potassium excretion in the distal tubulus and in comparison to the sodium re-absorption, an estimate of aldosterone activity in serum. This is also backed up by the increased activity of aldosterone post race. In the first two days, the potassium-to-sodium ratio was > 1.0 and we must assume that aldosterone activity was higher in these days compared to the rest of the race since the RAAS-axis is

stimulated in an intensity-dependent fashion.²⁵ This might also be backed up with the faster stages times in the beginning of the race and the finding, that both post race aldosterone and the change in aldosterone were not related to both the post race potassium-to-sodium ratio and the change in potassium-to-sodium ratio in urine. The timeframe over ten days was probably too long that aldosterone would have been increased highly enough.

A second hypothesis was that the increase in aldosterone would be related to the increase in extracellular water. Although extracellular water increased, the increase in extracellular water was neither related to both post race serum aldosterone nor to the change in serum aldosterone. Speedy *et al.* measured both vasopressin and aldosterone in five subjects and 11 controls after an Ironman triathlon.⁴ The median post race aldosterone in the subjects did not differ significantly from the controls. They found no evidence that aldosterone contributed to fluid retention. Regarding recent findings, an increased activity of vasopressin may contribute to the pathogenesis of EAH.^{7,8} However, vasopressin showed no change in this race and the increase in extracellular water was neither related to post race vasopressin nor to the change in vasopressin. Likewise, vasopressin and urinary osmolality showed no relationship. Speedy *et al.* measured vasopressin in addition to aldosterone in their subjects and controls after an Ironman triathlon.⁴ The median post race vasopressin was significantly lower in the subjects compared to the controls. They concluded that fluid retention was not due to vasopressin. Also, already 30 years ago, Williams *et al.* found during a multi-stage performance an increase in plasma volume, but no increase in vasopressin.¹⁴ One problem in the determination of the activity of vasopressin during a field study is its short half life time of 24.1 min.²⁶ Considering that the athletes got slower and slower during the race, the activity of vasopressin will also be reduced. The fastest Ironman was on Day 1, the slowest Ironman was on Day 9. This might also explain why vasopressin was not increased post race since vasopressin release is intensity dependent.²⁷ There is also a delay between the arrival at the

finish line and the blood sampling under field conditions.⁴ The determination of Copeptin as a stable peptide and precursor of vasopressin might be more useful to detect hormonal changes.²⁸⁻³⁰

Another problem regarding the determination of the fluid retention could be the applied method of bioelectrical impedance analysis. We measured total body water, extracellular water and intracellular water using the bioelectrical impedance analysis method. This method is reliable under both laboratory situations²² and field conditions.⁹ However, hydration status²³ and extracellular osmolality²⁴ must be stable. In our athletes, urinary specific gravity, urinary osmolality and urinary sodium remained unchanged throughout the race. Since urinary indices are reliable indices of hydration status³¹ with urine osmolality as the most promising marker available,³² we consider the results of bioelectrical impedance analysis for both total body water and extracellular water reliable. In a recent study in a multi-stage mountain bike race, the time course of the decrease in body mass and the increase in extracellular water could be described in the same manner using the same method.¹¹ In another recent study in a multi-stage cycling race, an increase in lean body mass was described while using dual energy x-ray absorptiometry.³³ The meaning of this increase in lean body mass remains unclear, and the development of an oedema might be postulated.³⁴ However, we found no association between the increase in lean body mass and the increase in both total body water ($r = -0.36, p = 0.33$) and extracellular water ($r = -0.36, p = 0.33$).

One might assume that the non-finishers may have dropped out due to disorders in fluid and electrolyte metabolism since EAH in Ironman triathletes may amount to ~18%.³⁵ However, EAH in Ironman triathletes is usually asymptomatic³⁵ and the non-finishers in this race gave up due to accident (1), overuse injuries of the lower limbs (1), fatigue and overuse injuries of

the lower limbs (2), respiratory problems (1), and loss of motivation (1). These symptoms were not typical for EAH.⁵

This investigation is limited in that haemoglobin, haematocrit, plasma sodium and plasma potassium were not determined before and after each stage in order to show changes between performance and recovery. Also the data of fluid intake and urinary output would give more insight into fluid regulation.¹¹ In athletes at the edge of physical performance and without a support crew; however, it is not possible to burden them unnecessarily. The non-invasive investigation such as measuring body composition using bioelectrical impedance analysis² and urinary analysis³⁶ takes little time per day, does not bother ultra-endurance athletes while racing and resting, and provides reliable data. However, analysis of haemoglobin, haematocrit and plasma electrolytes could be performed in ultra-endurance athletes using a point-of-care (POC) analyzer such as the i-STAT,³⁷ where only capillary samples would be needed.³⁸

To summarise, we found an increase in aldosterone and extracellular water after this multi-stage ultra-endurance triathlon. However, we could not establish an association between the increase in extracellular water and the increase in aldosterone. Furthermore, we found neither an increase in vasopressin, nor an association between both the change in vasopressin and post race vasopressin with the increase in extracellular water. **The increase in total body water and extracellular water could not be explained by hormonal changes.** In future studies, the marker Copeptin should be determined as a stable peptide and precursor of vasopressin that might be more useful to detect hormonal changes during ultra-endurance performance and a potential association between the increase in extracellular water and hormonal changes. **Renal function might have changed during these 10 days and excretion of water might have been reduced. Furthermore, intake of fluid during the heat of the day might have led to fluid overload.**

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Day	Finish time (min)	Temperature (°C)
1	760 (106)	27
2	849 (166)	26
3	815 (95)	28
4	855 (108)	27
5	897 (133)	29
6	882 (127)	31
7	906 (119)	31
8	889 (139)	23
9	953 (155)	19
10	940 (203)	23

Table 1: Daily finish times per Ironman distance and daily highest temperature. Race times are presented as mean (SD).

Parameter	Before stage 1 (n=5)	After stage 1 (n=5)	Before stage 2 (n=4)	After stage 2 (n=4)	Before stage 3 (n=4)	After stage 3 (n=4)
Body mass (kg)	76.6 (5.7)	72.7 (4.9) ^{§,*}	74.0 (5.2) [§]	72.8 (5.1) ^{§,*}	73.1 (4.6) ^{§,*}	72.2 (4.6) [§]
Fat mass (kg)	11.1 (2.7)	8.6 (1.9)	10.7 (1.9) [§]	8.8 (1.0) [§]	10.7 (0.9) ^{§,*}	9.7 (1.3) ^{§,*}
Lean body mass (kg)	65.6 (5.0)	64.1 (3.3)	63.3 (5.1)	64.0 (4.6)	62.4 (5.1)	62.6 (5.3)
Protein mass (kg)	13.1 (0.9)	12.9 (0.7)	12.8 (1.0)	12.9 (1.0)	12.5 (1.0)	12.5 (1.0)
Total body water (L)	48.4 (3.8)	47.2 (2.5)	46.5 (3.8)	47.1 (3.5)	46.0 (3.8)	46.1 (4.0)
Extracellular water (L)	15.6 (1.5)	14.8 (0.9)	14.5 (1.4)	14.7 (1.2)	14.7 (1.4)	14.8 (1.5)
Intracellular water (L)	32.8 (2.3)	32.4 (1.7)	32.0 (2.5)	32.4 (2.4)	31.3 (2.4)	31.3 (2.5)
Urinary sodium (mg)	176.4 (43.1)	95.8 (46.0)	83.4 (30.5)	82.8 (15.7)	102.5 (41.0)	116.3 (44.4)
Urinary potassium (mg)	73.6 (72.9)	132.4 (57.3)	51.5 (14.8)	135.1 (51.8)	43.1 (11.4)	137.2 (38.7)
Potassium-to-sodium ratio	0.50 (0.65)	1.61 (0.92)	0.70 (0.40)	1.60 (0.43) [§]	0.49 (0.27) [*]	1.36 (0.75)
Urinary specific gravity (g/mL)	1.016 (0.004)	1.028 (0.004)	1.025 (0.004)	1.030 (0.001)	1.034 (0.004)	1.022 (0.010)
Urinary osmolality (mosmoL/kg)	746.3 (146.4)	940.4 (177.3)	909.5 (134.9)	1,086.3 (203.4)	1,016.9 (179.7)	1,138.1 (143.0)

Table 2: Results of the non-finishers were only included when an athlete finished the stage and results from both pre and post stage were available. Results are expressed as mean (SD). [§] = $p < 0.05$ versus baseline value. ^{*} = $p < 0.05$ versus previous value.

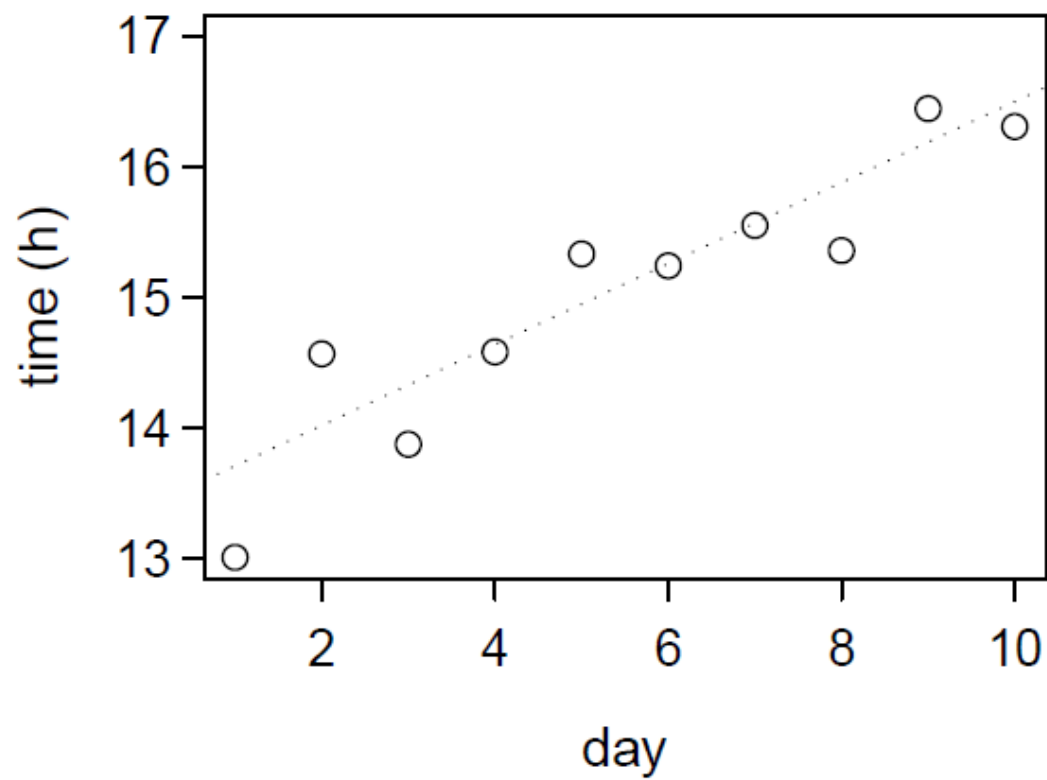


Figure 1: Daily finish times throughout the race. Time per Ironman increased significantly throughout the race ($p < 0.0001$).

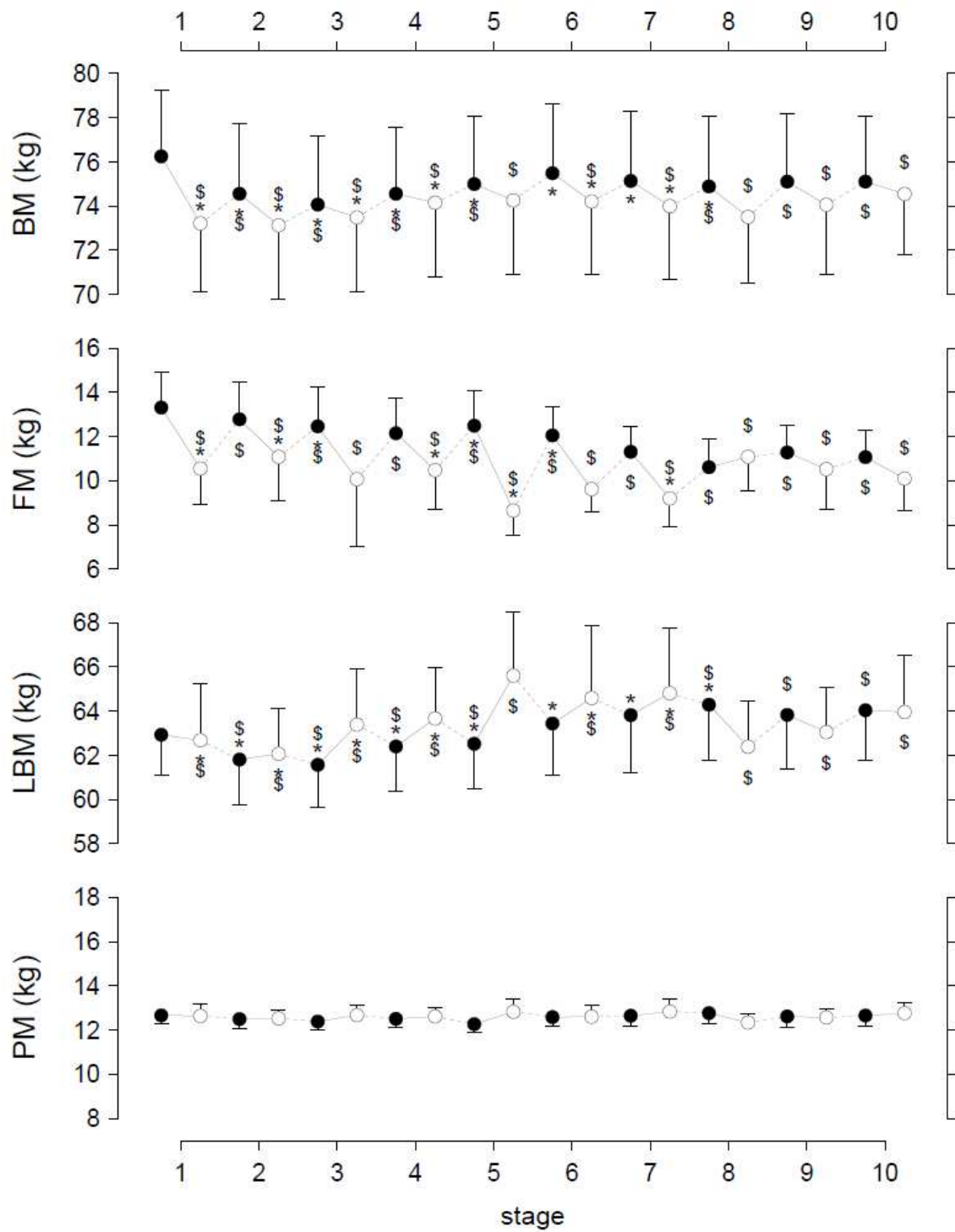


Figure 2: Changes in body mass (BM), fat mass (FM), lean body mass (LBM) and protein mass (PM) during the 10 days. \$ = $p < 0.05$ versus baseline value. * = $p < 0.05$ versus previous value.

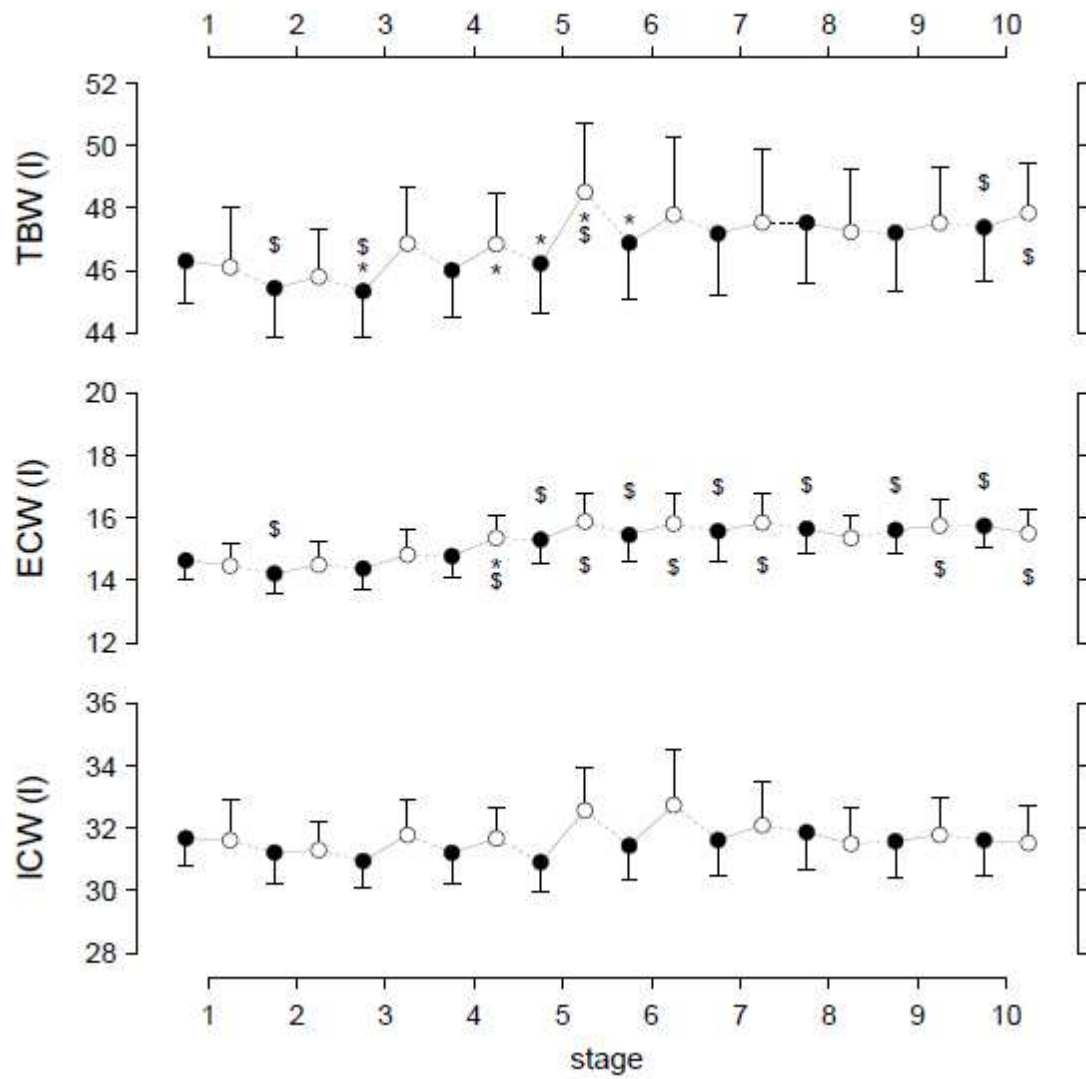


Figure 3: Changes in total body water (TBW), extracellular water (ECW) and intracellular water (ICW) during the 10 stages. \$ = $p < 0.05$ versus baseline value. * = $p < 0.05$ versus previous value.

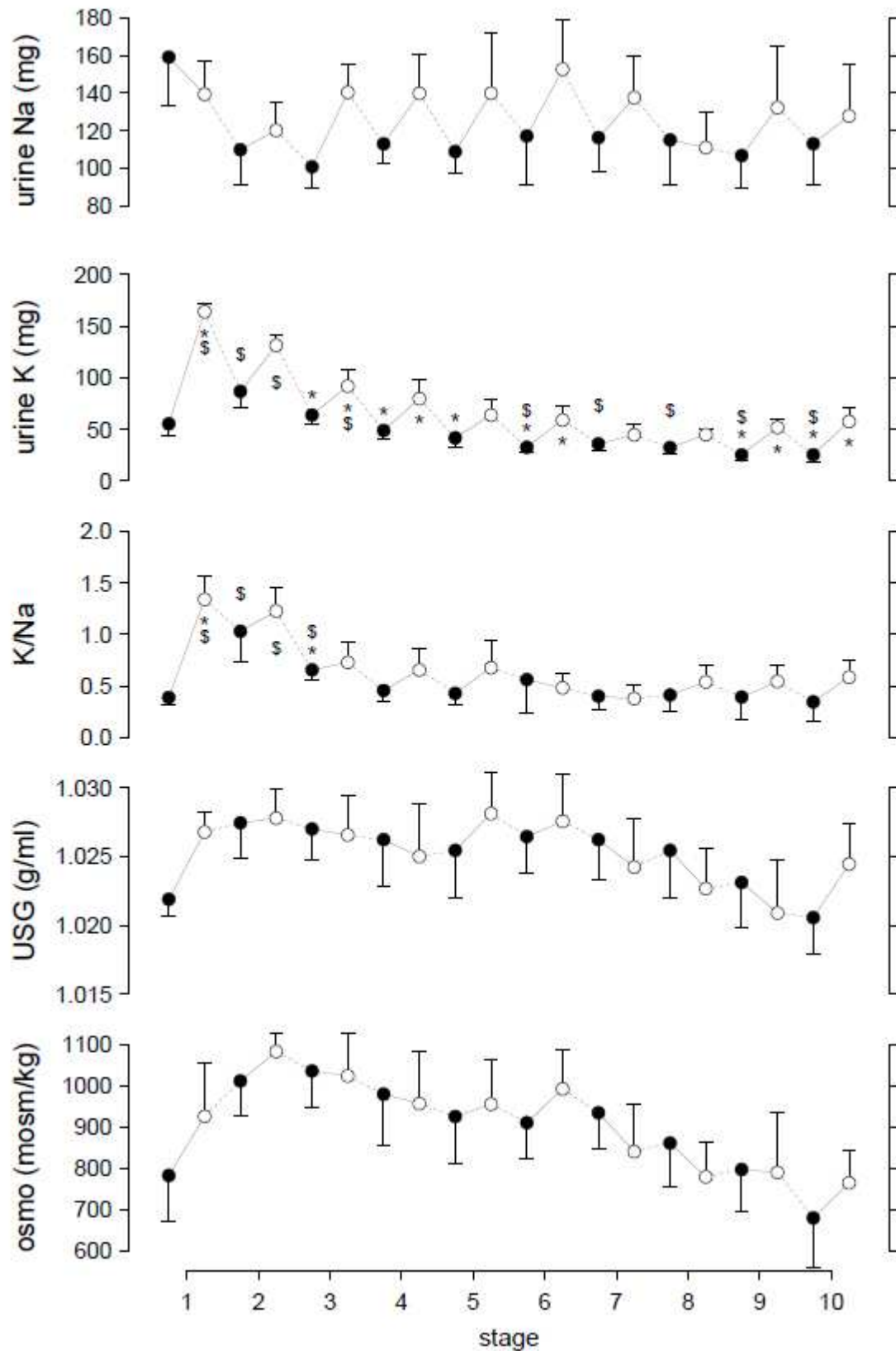


Figure 4: Changes in urinary sodium (urine Na), urinary potassium (urine K), the potassium-to-sodium ratio (K/Na), urinary specific gravity (USG) and urinary osmolality (osmo) during the 10 days. \$ = $p < 0.05$ versus baseline value. * = $p < 0.05$ versus previous value.